

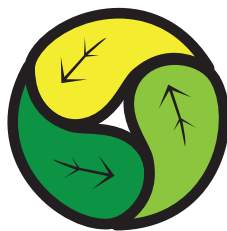
**УНИВЕРЗИТЕТ „ГОЦЕ ДЕЛЧЕВ” – ШТИП**  
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Оригинален научен труд

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## СЕНЗОРНА И АНАЛИТИЧКА ЕВАЛУАЦИЈА НА ЛАДНО-ЦЕДЕНИ МАСЛА ОД СОНЧОГЛЕД

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Љупчо Михајлов<sup>1</sup>

**Апстракт:** Составот на масните киселини, испарливите компоненти и сензорна евалуација на 16 ладноцедени сончогледови масла беа предмет на ова истражување. Низок удел на мононезаситена олеинска киселина и висок удел на полинезаситена линолеинска киселина (со удел преку 60%) може да предизвика брза оксидација на маслото во споредба со масла кои имаат висок удел на олеинска киселина и да предизвикаат „ужегнат“, „кисел“ или „горчлив“ вкус. Количеството на  $\alpha$ -пинен може да служи како маркер при процесот на производство на ладноцедено сончогледово масло. Овој монотерпен е многу испарлив. Според тоа, екстремно ниски концентрации на овој монотерпен, како и други испитувани монотерпени укажуваат на незадоволителен квалитет на сончогледови зрна или несоодветен третман на зрната пред процесот на ладно цедење.

**Клучни зборови:** ладноцедени сончогледови масла, испарливи компоненти, состав на масни киселини, сензорна евалуација.

## SENSORY AND ANALYTIC EVALUATION OF COLD-PRESSED SUNFLOWER OILS

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Ljupco Mihajlov<sup>2</sup>

**Abstract:** Fatty acid composition, volatile profile and sensory evaluation of 16 cold pressed sunflower oils were object of this study. Low abundance of monosaturated oleic acid and very high abundance of polyunsaturated linoleic fatty acid (presented in abundance more than 60%) can cause faster oxidation in comparison to high oleic sunflower oils which can induce unpleasant ‘rancid’, ‘sour’ or ‘bitter’ taste. The quantity of  $\alpha$ -pinene can be used as also marker for the process of cold-pressing of sunflower oil. This monoterpene hydrocarbon is very volatile and extremely low level of  $\alpha$ -pinene as well as significant lower concentration of all other monoterpenes can lead us to conclusion that the quality of the seeds is very poor or incorrect pretreatment of the seeds was applied.

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**Key words:** *cold-pressed sunflower oils, volatile compounds, fatty acid composition, sensory evaluation*

## 1. Introduction

Vegetable oils are divided in two types. The first one belongs to the oils in terms of energy storage in plants as fatty acids bond in triglycerides, typical for oils derived from different plant seeds. The second one belongs to essential oils which are produced as self-defense or attractant compounds. Most of essential oils are used in flavour industries such as food, beverage, traditional medicine and perfumery because of their terpenic nature. Over 95% of the *Citrus* essential oils are consisted from monoterpenes and sesquiterpenes with domination of limonene as main monoterpene hydrocarbon [1].

Virgin sunflower oil seems to be link between those two types of oil. The sunflower seeds contain fixed oil as the major oil which is mixture of saturated and unsaturated fatty acids mainly bonded as triglycerides and minor quantity of terpenic oil with predominant fraction of monoterpene hydrocarbons [2].

Virgin oils are very popular foodstuff as a 100% natural oils produced only with cold pressing without any step of rafination [3]. Edible oils produced on this way usually had higher concentrations of antioxidant compounds such tocopherols, carotenoids and chlorophylls and stronger impact on human health in comparison to refined oils [4, 5].

It is very well known positive relation between edible oil consumption and reduced risk of coronary hearth diseases, the level of LDL, degenerative diseases and cancer [6]. However, except essential fatty acids and Vitamin E, the chemical composition of edible oils from different plants represents minor grope of phenolic components as special grope of powerful antioxidants responsible for human health benefits [7].

Traditional Balkain cuisine, as well as the other cuisines of South-East European countries, includes sunflower oil as the most used cold-pressed and refined oil for cooking and frying. It has excellent nutritional properties, due to the high level of  $\alpha$ -tocopherol, oleic and linoleic fatty acids. Also, this oil is favorable because of very low level of saturated fatty acids (palmitic and stearic acid) with percentages less than 15%. Abundance of oleic and linoleic acids divide the sunflower oils on two types: high oleic sunflower oils with percentage of oleic acid around 60% and high linoleic sunflower oils with abundance of linoleic acid higher than 80% [8].

However, the most important parameter for the evaluation of the quality of virgin oils is the sensory impression, because more than any other parameter, the appearance and the taste of a product deeply influence the buying decision of the consumer. Cold pressed sunflower oil with acceptable quality has to be



clear oil with pale yellow colour. Typical attributes are sunflower-seed like, terpenic like, citrus like, nutty and sometimes fruity for virgin sunflower oil. Unpleasant flavor of defects in the taste of the oils is usually described as rancid, bitter and sour taste [9].

It is obvious that there are many published papers about volatile compounds in sunflower oils but they are always linked to degree of oxidation [11].

Only few published papers are connected to the volatile fraction of sunflower oil. In the work of Cioni *et al.* the percentages of  $\alpha$ -pinene in two cultivars Carlos and Florom 350 were 53.6 and 43.1 respectively following by other monoterpenes such sabinene,  $\beta$ -pinene and limonene. According to their findings, the fatty acid profile of the cultivar Carlos was dominant linoleic acid with percentage of 58% and the Florom 350 cultivar was dominant oleic acid with percentage of 50.1% [12]. In the work of Krist *at al.*,  $\alpha$ -pinene was used as a marker of adulteration of poppy seed oil with sunflower oil [13].

Oxidation of fatty acid in sunflower oil was object of study in the work of Kockritz and Martin [14]. According to their results, abundance of linoleic acid in high linoleic sunflower oils was in range of 48.3-74 % and abundance of oleic acid in high oleic sunflower oils was in range of 91-92 % [14].

To the best of our knowledge, there are only few published studies on the volatile profile of cold-pressed sunflower oil. At this point of view, this work had two aims. The first aim of this manuscript is to find possible relationship between quantities of five monoterpenes ( $\alpha$ -pinene,  $\beta$ -pienen, limonene, champhene and  $\alpha$ -phelandrene) and sensory evaluation of the oils and the second aim of the work is to explain how percentages of fatty acid and abundance of monoterpenes can be used as markers of the quality of the seeds from sunflower.

## **2. Materials and Methods**

### **2.1. Samples**

The first thirteen samples of cold-pressed sunflower oils were commercial oils. Sunflower oils 15 and 16 were cold-pressed in laboratory from the selected seeds with typical musty and fasty flavour.

### **2.2. Reagents and standards**

The standards of monoterpenes  $\alpha$ -pinene,  $\beta$ -pienen, limonene, champhene and  $\alpha$ -phelandrene were purchased from Merck, Germany. SPME cartridge 65  $\mu$ m PDMS/DVB, 50/30  $\mu$ m CRB-DVB-PDMS, 85  $\mu$ m were purchased from Sigma Aldrich, Belgium.



### **2.3. Determination of fatty acid composition**

The fatty acid composition of cold pressed sunflower oils was determined using chiral gas chromatography. The esters were prepared using 2 drops of each oil dissolved in 1 ml of heptane. After addition of 50  $\mu\text{L}$  of sodium methylate with concentration of 2 mol/L, the samples were homogenized. After homogenization, 100  $\mu\text{L}$  of distilled water was added in each sample. Samples were centrifuged and two phases was appeared. The down phase was removed and upper phase was dissolved in 50  $\mu\text{L}$  of 1 M HCl. After second homogenization, the red colour was detected and sodium sulphate anhydride was added in order to remove the water traces. Finally, upper phase was transferred in GC vials and fatty acid methyl esters were analyzed. Fatty acid methyl esters (FAME) were analyzed using a capillary GC equipped with a CP7420 Select FAME column, 100 m x 0.25 mm internal diameter with 0.25  $\mu\text{m}$  film thickness. Analyzes were performed on Agilent 6890 equipped with KAS4Plus and FID. The oven temperature was programmed to increase from 150°C to 240°C with rate of 1.5°C/min and maintained isothermal at 240°C 20 min. The injector and detector temperature were both 260°C. Hydrogen was used as the carrier gas at an average velocity of 25 ml/min. The retention times of separated picks were confirmed by FAME standards.

### **2.4. The extraction of volatile compounds by HS-SPME**

The extraction of free volatiles was performed by application of solid phase microextraction in head-space mode (HS-SPME). Regarding selection of the fibers, the 50/30  $\mu\text{m}$  Carbowax-Divinylbenzene-Polydimethylsiloxan (CRB-DVB-PDMS) fiber was more sensitive than the 85  $\mu\text{m}$  polyacrylate fiber. The contact time between fiber and head-space was 30 min. The most appropriate extraction temperature was 45°C. All analyses were performed in triplicate.

### **2.5. Identification and quantification of monoterpenes in sunflower oils**

The volatile compounds of cold-pressed sunflower oils were analyzed using a HP 6890 GC equipped with a single quad Mass Spectrometer (MS) HP5973 and HP Carbowax cross-linked fused-silica capillary column (i.d. = 0.25 mm, length = 60 m, film thickness = 0.25  $\mu\text{m}$ ). The interface temperature was 280°C; Injector and detector temperatures were set at 260°C, respectively. Acquisition mass range was 40-400m/z and solvent cut 2min. Flame ionization detector was used for quantification of volatile compounds. The temperature gradient was 45°C to 180°C at 3°C/min and then from 180°C to 260°C; Helium was the carrier gas at 35kPa (32 cm/s).

The components of the sunflower oil were identified by comparison of their linear retention indices, with those from pure standards or reported in



literature. Comparison of fragmentation patterns in the mass spectra with those stored on databases and MS data of our collection was also performed.

Quantification of  $\alpha$ -pinene,  $\beta$ -pinene, limonene,  $\alpha$ -phellandrene and champene was performed with external calibration using a standard solution of terpenes (Table 1). Standard solutions used for calibration were prepared in refined sunflower oil with stock solution of 80 mg of monoterpene in 80 mg of refined oil. The standards was mixed with the oil overnight and kept in the dark bottles at room temperature. The calibration curves were established by plotting the area of peaks against different concentrations of monoterpenes with correlation factor given in Table 1.

**Table 1.** Calibration curve of pure standards

Standard of terpenes	Calibration curve	R <sup>2</sup>
$\alpha$ -pinene	$y=224393x-132250$	0.9973
$\beta$ -pinene	$y=95281x+103378$	0.9961
$\alpha$ -phelandrene	$y=79419x+54734$	0.9992
limonene	$y=130416x-52401$	0.9936
camphene	$y=96739x-56442$	0.9985

## 2.6. Sensory evaluation of virgin sunflower oils

According to the standards for sensory evaluation of the working grope of Dr. Mathäus *et al.*, the assessment a scale from 0 (not detectable) to 5 (very strongly perceivable at the level of saturation) was used. Virgin sunflower oil produced from the dehulled seeds with high-quality had very pleasant ‘sunflower seed-like’, ‘nutty’ and sometimes ‘fruity’ taste. According to their opinion, oils from whole seeds have a stronger taste and especially the sensory attributes wood-like, astringent and bitter are predominant, which was not an unpleasant or ‘off’ taste of virgin sunflower oil. On this way, the whole taste and smell of sunflower oils were estimated. However, very frequent unpleasant sensory attributes are described as ‘rancid’, sour ‘fusty’ or ‘musty’. These attributes should not be present in virgin sunflower oil with high quality because the presence of these attributes indicates to failures during storage of the raw material (fusty, musty), during processing (roasty, burnt) or during storage of the oil (rancid).

## 2.7. Statistical analyses

One-way ANOVA and Principal component analysis (PCA) was performed to gain an overview of how the samples were correlated to each other with regard to equilibrium volatile headspace concentration and fatty acid percentages. Correlation matrix was applied in multivariate analysis with





Minitab software release 14 so that the data was auto scaled by variable to give the same weight to all components.

### 3. Results and discussion

Fatty acid composition of the oils is presented in Table 2. Sunflower oils 1 and 10 had the lowest percentage of palmitic acid (3.90%) and the highest percentage had the sunflower oil 3 (7.10 %). Also, sunflower oil 1 and 10 are high-oleic oils with abundance of 83.35% and 81.82% respectively. The abundance of stearic acid in all 16 samples of sunflower oils was in the range between 4.38% and 6.30%. Behenic acid was present in percentages less than 1% in all 16 samples of sunflower oils. All other samples belongs to the high linolenic type of oils with percentage of linolenic acid between 52.32 and 65.61%.

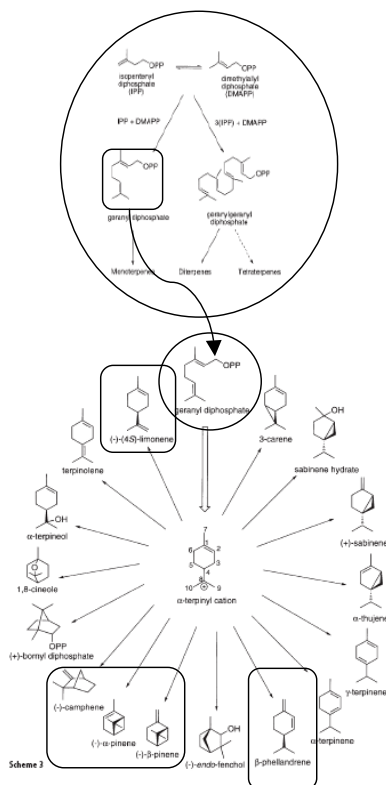
**Table 2.** Fatty acid composition of cold pressed edible oils (%) and relative standard deviations (n=3) of virgin cold pressed sunflower oils

	C16:0	C18:0	C18:1D9	C18:1D11	C18:2	C22:0
Sunflower oil 1	3.90±0.00	4.38±0.05	1.01±0.05	<b>83.35±0.22</b>	<b>5.61±0.01</b>	0.74±0.05
Sunflower oil 2	5.31±0.26	5.21±0.23	0.59±0.15	31.15±0.08	55.98±0.57	0.69±0.09
Sunflower oil 3	<b>7.01±0.01</b>	4.65±0.11	0.74±0.27	25.90±0.11	60.02±0.07	0.54±0.03
Sunflower oil 4	6.71±0.10	5.45±0.00	0.56±0.22	19.97±0.20	65.61±0.01	0.66±0.00
Sunflower oil 5	6.78±0.00	5.31±0.02	0.57±0.18	<b>19.89±0.01</b>	65.91±0.01	0.69±0.00
Sunflower oil 6	6.48±0.07	5.28±0.00	0.85±0.01	24.77±0.03	61.07±0.00	0.66±0.05
Sunflower oil 7	6.53±0.00	5.37±0.04	0.81±0.03	24.87±0.11	60.84±0.14	0.58±0.04
Sunflower oil 8	5.77±0.02	5.66±0.01	0.71±0.06	27.97±0.11	58.67±0.17	0.51±0.01
Sunflower oil 9	6.30±0.00	<b>6.30±0.03</b>	0.63±0.03	27.24±0.03	58.19±0.07	0.52±0.00
Sunflower oil 10	<b>3.89±0.08</b>	4.65±0.03	1.01±0.05	<b>81.82±0.31</b>	<b>7.02±0.26</b>	0.71±0.06
Sunflower oil 11	6.54±0.08	5.69±0.05	0.82±0.03	26.06±0.12	59.44±0.13	0.57±0.00
Sunflower oil 12	6.50±0.18	5.38±0.11	0.98±0.02	25.74±0.12	60.49±0.15	0.52±0.05
Sunflower oil 13	6.39±0.15	5.20±0.08	0.56±0.06	26.76±0.41	59.91±0.07	0.51±0.03
Sunflower oil 14	6.00±0.05	5.07±0.00	0.54±0.05	24.35±0.77	63.02±0.20	0.42±0.02
Sunflower oil 15	6.58±0.09	4.50±0.09	1.02±0.04	33.31±0.13	53.46±0.07	0.61±0.05
Sunflower oil 16	6.76±0.04	4.59±0.01	1.24±0.16	33.91±0.00	52.32±0.19	0.56±0.01

Terpenes as major components of plant essential oils are derived from mevalonic acid pathway. In brief, mevalonic acid is formed by condensation of acetyl-CoA and acetoacetyl-CoA. At the first step is formed 3-hydroxy-3-methylglutaryl-CoA (HMG-CoA) which is catalyzed by the enzyme HMG-CoA synthase. In the next step, HMG-CoA reductase (HMGR) catalyses deacylation of HMG-CoA to mevalonate (MVA). Mevalonic acid is converted



to geranylpyrophosphate, the most important precursor of monoterpene biosynthesis pathway. Then, geranylpyrophosphate is further modified to form various monoterpenes and other larger terpenoids (Scheme 1).



**Scheme 1.** Biosynthetic pathway of  $\alpha$ -pinene,  $\beta$ -pinene, limonene,  $\alpha$ -phellandrene and champene

According to the published results,  $\alpha$ -pinene,  $\beta$ -pinene, limonene,  $\alpha$ -phellandrene and champene are the most important monoterpenes, responsible for characteristic citrus-like and terpenic-like flavour of many essential oils [9, 10]. Results from the quantity of those monoterpenes given in Table 3 are in very good correlation with the results in the work of Bocci *et al.* which explain that more than 84% of all volatile compounds belong to  $\alpha$ -pinene. Concentrations of  $\beta$ -pinene, limonene,  $\alpha$ -phellandrene and champene are also in very good correlation with results from the work of Bocci *et al.* [9]

Many published manuscripts had confused results for aroma activity of monoterpenes in food. One part of our work was to estimate the contribution of



each monoterpene in the overall flavor of sunflower oil by gas-chromatography-olfactometry and sniffing of each compound after its elution from the sniffing port of the FID detector. Although  $\alpha$ -pinene is the major monoterpene in sunflower oil, its aroma potential is not very low and its contribution to the overall flavor of the sunflower oil is minor. In the work of Rychlik *et al.*, odor threshold of  $\alpha$ -pinene in the air is between 18-35 ng/L with typical 'terpeny' note which in oils can be recognized as 'bitter' taste. However, during its elution, we consider very weak fresh smell which was minor in comparison to the quantity of this monoterpene in the oil.  $\beta$ -pinene is abundant in much lower concentration and also, its threshold in air is higher than 2000 ng/L. From this result and according to our examination in the range of elution of this monoterpene, we can conclude that  $\beta$ -pinene cannot contribute in overall flavour and the smell of sunflower oils.  $\alpha$ -phellandrene has much higher odor potential with threshold in air in the range between 40-93 ng/L. Our sniffing impressions in this range of elution estimated potent 'fresh', 'citrus' and 'floral' smell. This smell was more intense in sunflower oils 3, 8 and 16 with higher concentration of this terpene. Its concentration from 3.51 ppm to 10.19 ppm can be responsible for 'fruity' or 'floral' smell of this oil. Champhene had higher levels in the same samples including sunflower oil 9. However, during the elution of this monoterpene, we did not consider potent smell.

The highest abundances of all monoterpenes were presented in sunflower oil 3. Regarding the sensory point of view presented in Table 4, this oils had the highest values for assessment (4 for 'sunflower seed like' and 3 for 'woody like'). However, we cannot conclude that this sensory perception is correlate with the level of monoterpenes, because other sunflower oils had even better sensory properties (4 for 'sunflower seed like', 1 for 'woody like', 2 for 'sweet' and 'fruity like' and 2 for 'nutty') but, the levels of monoterpenes were not so high. It might be that process of cold-pressing and the quality of the seeds are key factors for the highest quantities of monoterpenes.

The worst taste had sunflower oil 15 with very sour taste (evaluation 4). Regarding the fatty acid profile and the levels of monoterpenes we cannot correlate this taste with some of measured values and probably this oil had failures during storage of the sunflower seeds. The most astringent oil was sunflower oil 4 and the strongest bitter sunflower oil was oil 16. Although many consumers detect the bitterness of the oils as bad taste, the bitterness and astringency can come from the cold-pressing of whole seeds without dehulling or if oils are reach with powerful antioxidant polyphenolic compounds which usually have bitter taste.



**Table 3.** Mean concentrations of free monoterpenes (ppm) and relative standard deviations (n=2) of virgin cold pressed sunflower oils

Sample	$\alpha$ -pinene	Limonene	$\beta$ -pinene	$\alpha$ -phellandrene	Camphene
sunflower oil 1	50.73 $\pm$ 1.31	1.05 $\pm$ 0.03	1.73 $\pm$ 0.31	1.90 $\pm$ 0.10	2.35 $\pm$ 0.80
sunflower oil 2	98.93 $\pm$ 0.31	1.29 $\pm$ 0.08	2.93 $\pm$ 0.08	2.40 $\pm$ 0.10	2.64 $\pm$ 0.09
sunflower oil 3	<b>227.01<math>\pm</math>11.63</b>	2.54 $\pm$ 0.01	3.86 $\pm$ 0.62	1.19 $\pm$ 0.23	1.17 $\pm$ 0.12
sunflower oil 4	51.46 $\pm$ 0.12	0.73 $\pm$ 0.13	3.51 $\pm$ 0.29	1.61 $\pm$ 0.02	1.80 $\pm$ 0.01
sunflower oil 5	52.96 $\pm$ 0.78	0.59 $\pm$ 0.01	1.86 $\pm$ 0.09	1.71 $\pm$ 0.02	1.13 $\pm$ 0.06
sunflower oil 6	37.31 $\pm$ 1.28	0.88 $\pm$ 0.04	2.60 $\pm$ 0.01	2.33 $\pm$ 0.02	2.64 $\pm$ 0.03
sunflower oil 7	<b>13.39<math>\pm</math>0.14</b>	0.61 $\pm$ 0.03	<b>0.53<math>\pm</math>0.01</b>	1.60 $\pm$ 0.02	1.37 $\pm$ 0.02
sunflower oil 8	90.99 $\pm$ 1.85	1.01 $\pm$ 0.02	8.10 $\pm$ 0.18	<b>4.97<math>\pm</math>0.13</b>	<b>5.54<math>\pm</math>0.10</b>
sunflower oil 9	<b>16.92<math>\pm</math>0.63</b>	1.07 $\pm$ 0.04	2.34 $\pm$ 0.05	1.37 $\pm$ 0.05	<b>7.06<math>\pm</math>0.08</b>
sunflower oil 10	54.15 $\pm$ 0.98	0.88 $\pm$ 0.03	4.38 $\pm$ 0.03	1.58 $\pm$ 0.01	1.48 $\pm$ 0.01
sunflower oil 11	36.13 $\pm$ 0.90	0.74 $\pm$ 0.00	2.22 $\pm$ 0.00	1.13 $\pm$ 0.00	1.58 $\pm$ 0.00
sunflower oil 12	40.37 $\pm$ 0.91	0.71 $\pm$ 0.00	1.84 $\pm$ 0.02	0.88 $\pm$ 0.00	1.52 $\pm$ 0.00
sunflower oil 13	38.39 $\pm$ 1.15	0.71 $\pm$ 0.00	1.90 $\pm$ 0.01	1.08 $\pm$ 0.01	1.51 $\pm$ 0.02
sunflower oil 14	<b>4.82<math>\pm</math>0.45</b>	<b>0.42<math>\pm</math>0.00</b>	<b>0.30<math>\pm</math>0.01</b>	<b>0.04<math>\pm</math>0.00</b>	<b>0.62<math>\pm</math>0.04</b>
sunflower oil 15	52.47 $\pm$ 0.27	0.70 $\pm$ 0.01	3.13 $\pm$ 0.04	2.17 $\pm$ 0.00	2.93 $\pm$ 0.00
sunflower oil 16	52.58 $\pm$ 0.58	1.09 $\pm$ 0.02	5.09 $\pm$ 0.05	<b>3.51<math>\pm</math>0.06</b>	<b>4.69<math>\pm</math>0.09</b>

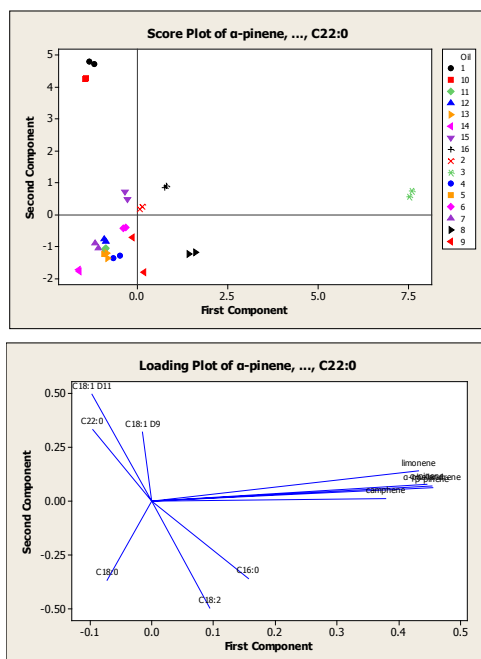
**Table 4.** Sensory evaluation of the taste of cold pressed sunflower oils (Panelist 1 and 2)

	Sunflower seed like	Nutty	Fruity, Sweet	Woody like	Astringent	Bitter	Rancid	Sour
Sunflower oil 1	3	1	2	0	0	0	0	0
Sunflower oil 2	2	2	2	1	0	0	0	0
Sunflower oil 3	2	1	1	3	0	0	0	0
Sunflower oil 4	3	1	1	4	3	2	0	0
Sunflower oil 5	<b>3</b>	<b>2</b>	<b>2</b>	<b>2</b>	<b>1</b>	<b>1</b>	<b>0</b>	<b>0</b>
Sunflower oil 6	2	1	2	1	1	0	0	0
Sunflower oil 7	2	1	2	1	1	0	0	0
Sunflower oil 8	1	1	1	1	1	1	1	1
Sunflower oil 9	1	1	1	1	1	1	1	1
Sunflower oil 10	1	1	0	1	1	0	0	0
Sunflower oil 11	0	0	0	1	1	2	1	0
Sunflower oil 12	0	1	0	0	0	0	0	0
Sunflower oil 13	1	1	0	0	0	0	0	0
Sunflower oil 14	0	0	0	1	1	1	2	0
Sunflower oil 15	0	0	0	2	2	4	0	4
Sunflower oil 16	1	1	0	3	2	3	0	0



**Table 4.** Sensory evaluation of the taste of cold pressed sunflower oils (Panelist 3 and 4)

	Sunflower seed like	Nutty	Fruity, Sweet	Woody like	Astringent	Bitter	Rancid	Sour
Sunflower oil 1	3	1	2	0	0	0	0	0
Sunflower oil 2	2	1	1	0	0	0	0	0
Sunflower oil 3	2	1	0	1	0	0	0	2
Sunflower oil 4	3	1	0	4	2	1	0	0
Sunflower oil 5	3	1	2	2	2	1	0	0
Sunflower oil 6	2	1	1	1	0	0	0	0
Sunflower oil 7	2	1	1	1	0	0	0	0
Sunflower oil 8	2	0	0	1	1	0	2	1
Sunflower oil 9	2	1	2	0	0	0	0	0
Sunflower oil 10	2	1	0	2	2	0	0	0
Sunflower oil 11	1	0	1	1	0	0	1	1
Sunflower oil 12	1	0	0	1	1	0	0	1
Sunflower oil 13	1	0	0	1	1	0	1	0
Sunflower oil 14	0	0	0	1	1	1	2	0
Sunflower oil 15	1	0	0	3	3	3	0	3
Sunflower oil 16	2	1	0	3	2	3	0	0



**Figure 1.** PCA score and loading plots of monoterpenes and fatty acids in sunflower oils



PCA score plots were used to determine whether sixteen different cold-pressed sunflower oils could be grouped into different classes (Fig. 1). If we compare score plot and loading plot, we can conclude that only for sunflower oil 3 the differentiation or closeness between the volatile flavour compounds directed in positive side of PC 1 was dependent on their quantity values. All monoterpene hydrocarbons namely  $\alpha$ -pinene,  $\beta$ -pinene, limonene, camphene and  $\alpha$ -phellandrene, as volatile compounds, could be classified in one group in PC1, because the coefficients of these volatile compounds were the same positive sign located in PC 1 (Fig. 1). In most cases, the differentiation or closeness between the volatile flavour compounds directed in positive side of PC 1 was dependent on their quantities.

Sunflower oils 16 and 8 were differentiated by the quantity of C18:1 D9 and the quantities of camphene and  $\alpha$ -phellandrene, respectively. As we can see from the table 1, the sunflower oil 16 was differentiated because of the highest percentage of C18:1 D9 with value of  $1.24 \pm 0.16$ . On the other hand, sunflower oil 8 was differentiated because of very high concentrations of  $\alpha$ -phellandrene and camphene with values of  $4.97 \pm 0.13$  ppm and  $5.54 \pm 0.10$  ppm, respectively. Sunflower oils 1 and 10 were differentiated from the other oils and very close on PC 2 because of the highest level of oleic acid and we can classify these oils as high oleic sunflower oils. Also, these oils had lowest and almost identical percentages of palmitic acid. On the basis of their fatty acid profile we can expect that those oils should have the longest oxidation stability.

On the negative part of PC 2 was differentiated sunflower oil 14 because of the lowest percentage of Valeric acid and very low percentage of C18:1 D11 ( $24.35 \pm 0.77\%$ ). Regarding the sensory point of view, this oil is classified as oil with very poor taste and very strong 'fatty' and 'musty' taste. Two reasons can explain very poor taste and smell of this oil. The first and less important reason is very low concentration of all identified and quantified monoterpenes due to their low impact to the overall flavor of the sunflower oil. On the other hand, more important reason is very low abundance of oleic and very high abundance of linoleic fatty acid. Linoleic acid in this oil (presented as  $63.02 \pm 0.20\%$ ) can cause faster oxidation in comparison with high oleic sunflower oils which can induce unpleasant 'rancid', 'sour' or 'bitter' taste as was general opinion for the taste of this oil. Additionally explanation for very bad sensory characteristic of this oil is extremely low level of  $\alpha$ -pinene ( $4.82 \pm 0.45$  ppm) in comparison to the abundance of concentration of other four estimated monoterpenes.  $\alpha$ -pinene is also marker for the process of cold-pressing [9,10]. This monoterpene hydrocarbon is very volatile and during process of refinement is removed from the oil. Extremely low level of  $\alpha$ -pinene as well as significant lower concentration of all other monoterpenes can lead us



to conclusion that the quality of the seeds is very poor or incorrect pretreatment of the seeds was applied [9,10].

Regarding the bitter taste of the sunflower oil, samples from sunflower oil 15 and 16 had the highest grades (3 and 4) and very sour taste for sunflower oil 15. The strong sour taste cannot be result of the concentration of monoterpenes because the terpenic profile of these oils is very similar to profile of other oils which taste is not described as very sour. The reason might be poor quality of the seeds and laboratory production of the oils with cold-pressing without any stage of seeds pretreatment. Also, during laboratory screw pressing, the process of sedimentation of the oils was excluded. During the process of sedimentation, the waxes and other undesirable compounds responsible for bitter taste are still present in the oils and these compounds might be responsible for ‘bitter’ taste of the oils.

It seems that the best sensory appreciation of sunflower oil 1, 5 and 6 was not related neither by their fatty acid profile neither by their terpenic profile. If we compare results from Tables 1 and 3, we can notice that those oils had very similar and not favorite fatty acid profile with low level of oleic and high level of linoleic acid which can induce instability. Also, terpenic profile of those two oils is similar with higher concentration of  $\beta$ -pinene ( $3.51 \pm 0.20$  ppm) which is not aroma active. However, these oils had grade of 3 for ‘sunflower seed like’ and 4 for ‘woody like’ which can lead us to two conclusions. The first conclusion is connected to the seed pretreatment. Strong ‘woody’ note is usually connected to screw pressing of whole seeds without dehulling. The second conclusion is that neither of five monoterpenes was responsible for typical ‘sunflower-seed’ taste of sunflower oils.

#### 4. Conclusion

Fatty acid composition, volatile profile and sensory evaluation of cold pressed sunflower oils were object of this study. Although  $\alpha$ -pinene was the major monoterpene in all samples, its contribution to the overall taste of the oils is very small. The results from our study we can conclude that  $\beta$ -pinene also cannot contribute in overall flavour and the smell of sunflower oils.  $\alpha$ -phellandrene has much higher odor potential with threshold in air in the range between 40-93 ng/L and its presence in the oils indicated potent ‘fresh’, ‘citrus’ and ‘floral’ smell and taste. This smell was more intense in sunflower oils 3, 8 and 16 with higher concentration of this terpene. The oils which higher level of oleic acid was more stable against oxidation in comparison to high linoleic sunflower oils.



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